Cp 1811
1654
#17
Mrs Plunkett
9-17-98

K:\CEGNER\DUK\9603PA\DU9603PA.TCV
CAE35/WP7.0
DEB/CAE/ach
March 13, 1998

PATENT APPLICATION
DOCKET NO.: DUK96-03pA

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Jonathan S. Stamler
Serial No.: 08/616,371 Group Art Unit: 1811
Filed: March 15, 1996 Examiner: B. Celsa
For: METHODS FOR PRODUCING AND USING S-NITROSOHEMOGLOBINS

CERTIFICATE OF MAILING

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as First Class Mail in an envelope addressed to Assistant Commissioner for Patents, Washington, D.C. 20231

on 3/13/98 (Date) Anita Heckman (Signature)

Anita Heckman
Typed or printed name of person signing certificate

TRANSMITTAL OF EXHIBIT A

Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

Transmitted herewith is Exhibit A, an abbreviated curriculum vitae of Joseph Bonaventura, Ph.D., inadvertently omitted from the package containing a Supplemental Reply and a Declaration of Joseph Bonaventura, Ph.D. Under 37 C.F.R. § 1.132, which was mailed to the United States Patent and Trademark Office on March 12, 1998.

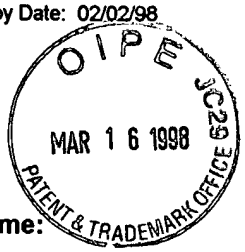
Respectfully submitted,

Carol A. Egner

Carol A. Egner
Registration No. 38,866
Attorney for Applicants
Telephone: (781) 861-6240
Facsimile: (781) 861-9540

Lexington, MA 02173
Dated: March 13, 1998

Copy Date: 02/02/98



JOSEPH BONAVENTURA

CURRICULUM VITAE

Name: Joseph Bonaventura, Ph.D.

Primary academic appointment: Nicholas School of the Environment
Secondary academic appointment: Department of Cell Biology, Duke Univ. Medical School
Present academic rank and title: Professor of Cell Biology

Date and rank of first Duke faculty appointment: 1972 - Associate

Social Security Number: 566-58-6023

Date of birth: February 15, 1942
Place of birth: Oakland, California
Citizenship: United States

Education:

1964 B.A., San Diego State College, California
1968 Ph.D., University of Texas, Austin, Texas
1969 NIH Postdoctoral Fellow, California Institute of Technology
1971 EMBO Course on Relaxation Kinetics, Stockheim, Germany
1971 American Cancer Society Postdoctoral Fellow, Rome, Italy
1972 EMBO Postdoctoral Fellow, Rome, Italy

Scholarly Societies:

American Association for the Advancement of Science
American Chemical Society
American Society of Biological Chemists
American Society of Zoologists
Basic Sciences Council of the American Heart Association
Biophysical Society
British Society of Chemical Industry
Sigma XI

Professional academic career:

1972-75 Associate (=Instructor), Department of Biochemistry, Duke University Medical Center and Duke University Marine Laboratory, Beaufort, North Carolina
1975-80 Established Investigator of the American Heart Association
1975-84 Assistant Medical Research Professor of Biochemistry, Duke University Medical Center and Duke University Marine Laboratory, Beaufort, North Carolina
1977- Editorial Board Member of HEMOGLOBIN, International Journal for Hemoglobin Research
1978-1994 Director, Duke University Marine Biomedical Center
1980- Advisory Board of MOLECULAR PHYSIOLOGY
1980-84 Science Advisory Board of National Public Radio

EXHIBIT
A

Professi nal academic career cont.:

1984-88	Associate Professor, Department of Physiology, Duke University Medical Center and Duke University Marine Laboratory, Beaufort, North Carolina
1984-	Toxicology Executive Committee, Duke University
1984-	Assistant Director for Marine Biomedical Programs, Duke University Marine Laboratory
1985-	Board of Directors, North Carolina Biotechnology Center
1988-90	Associate Professor, Department of Cell Biology, Duke University Medical Center and Duke University Marine Laboratory, Beaufort, North Carolina
1990-	Professor, Department of Cell Biology, Duke University Medical Center and Duke University Marine Laboratory, Beaufort, North Carolina

1. Refereed Journals and Chapters in Books

1967

Bonaventura, J., and A. Riggs. Polymerization of hemoglobins of mouse and man: structural basis. *Science*, 158:800-802.

1968

Bonaventura, J. and A. Riggs. Hemoglobin Kansas, a human hemoglobin with a neutral amino acid substitution and an abnormal oxygen equilibrium. *J. Biol. Chem.*, 243:980-991.

1969

Bonaventura, J. Polymeric hemoglobins of the house mouse (*Mus musculus* L.): Isolation of cysteinyl peptides. Biochem. *Genetics*, 3:239-247.

1972

Bonaventura, J. Functional properties of normal and abnormal hemoglobin. pp. 131-142. In: *Hemoglobinopathies*, G. Astaldi, et al., editors. Archivo casa sollievo della sofferenza. Fossia, Italy.

Bonaventura, J., C. Bonaventura, B. Giardina, E. Antonini and J. Wyman. Partial restoration of normal functional properties in carboxypeptidase A-digested hemoglobin. *Proc. Nat. Acad. Sci., U.S.*, 69:2174-2178.

Bonaventura, J., W.A. Schroeder and Suen Fang. Human erythrocyte catalase: an improved method of isolation and a re-evaluation of reported properties. *Arch. Biochem. Biophys.*, 150:606-617.

Brunori, M., J. Bonaventura, C. Bonaventura, E. Antonini and J. Wyman. Carbon monoxide binding by hemoglobin and myoglobin under photodissociating conditions. *Proc. Nat. Acad. Sci., U.S.*, 69:868-871.

1973

Barra, D., F. Bossa, J. Bonaventura and M. Brunori. Hemoglobin components from trout (*Salmo irideus*): determination of carboxyl and amino terminal sequences and their functional implications. *FEBS Lett.*, 35:151-154.

Bonaventura, C., J. Bonaventura, E. Antonini, M. Brunori and J. Wyman. Carbon monoxide binding by simple heme proteins under photodissociating conditions. *Biochemistry*, 12:3424-3428.

Bonaventura, J., and G.B. Kitto. Ligand-linked dissociation of some invertebrate hemoglobins. pp. 493-507. In: *Comparative Physiology*, L. Bolis, K. Schmidt-Nielsen and S.H.P. Maddrell, editors. North Holland, American Elsevier, Amsterdam.

Brunori, M., J. Bonaventura, C. Bonaventura, B. Giardina, F. Bossa and E. Antonini. Hemoglobins from trout: Structural and functional properties. *Mol. Cell. Biochem.*, 1:189-196.

Brunori, M., B. Giardina, J. Bonaventura, D. Barra and E. Antonini. Properties of fish hemoglobins: the hemoglobin system of trout (*Salmo irideus*). pp. 477-492. In: *Comparative Physiology*, L. Bolis, K. Schmidt-Nielsen and S.H.P. Maddress, editors. North Holland, American Elsevier, Amsterdam.

Wilson, M.T., M. Brunori, J. Bonaventura and C. Bonaventura. Effect of steady illumination of the binding of carbon monoxide by carboxymethylated cytochrome c. *Biochem. J.*, 131:863-865.

1974

Bonaventura, C., B. Sullivan and J. Bonaventura. The effect of pH and anions on the functional properties of hemoglobins from *Lemur fulvus fulvus*. *J. Biol. Chem.*, 249:3768-3775.

Bonaventura, C., B. Sullivan, J. Bonaventura and S. Bourne. CO binding by hemocyanins of *Limulus polyphemus*, *Busycon carica* and *Callinectes sapidus*. *Biochemistry*, I3:4787-4789.

Bonaventura, J., C. Bonaventura, G. Amiconi, E. Antonini and M. Brunori. Functional properties of hemoglobin Leiden ($Alpha_2^A beta_2$ 6 or 7 Glu deleted). *Arch. Biochem. Biophys.*, 161:328-332.

Bonaventura, J., C. Bonaventura, M. Brunori, B. Giardina, E. Antonini, F. Bossa and J. Wyman. Functional properties of carboxypeptidase-digested hemoglobins. *J. Mol. Biol.*, 82:499-511.

Bonaventura, J., C. Bonaventura and B. Sullivan. Urea tolerance as a molecular adaptation of elasmobranch hemoglobins. *Science*, 186:57-59.

Bonaventura, J., C. Bonaventura and B. Sullivan. Hemoglobin of the electric Atlantic torpedo: *Torpedo nobiliana*: a cooperative hemoglobin without Bohr effects. *Biochim. Biophys. Acta*, 371: 147-154.

Bonaventura, J., R.G. Gillen, and A. Riggs. The hemoglobin of the Crossopterygian fish, *Latimeria chalumnae* (Smith): Subunit structure and oxygen equilibria. *Arch. Biochem. Biophys.*, 163:728-734.

Chiancone, E., N. Anderson, E. Antonini, J. Bonaventura, C. Bonaventura, M. Brunori and C. Spagnuolo. Effect of heme and non-heme ligands on subunit dissociation of normal and carboxypepti-dase-digested hemoglobin. *J. Biol. Chem.*, 249:5689-5694.

Chiancone, E., J.E. Norne, J. Bonaventura, C. Bonaventura and S. Forsen. Nuclear magnetic resonance quadrupole relaxation study of chloride binding to hemoglobin Abruzzo ($alpha_2^A beta_2$ 143 His--Arg). *Biochim. Biophys. Acta*, 336:403-406.

Sullivan, B., J. Bonaventura and C. Bonaventura. Functional differences in the multiple hemocyanins of the horseshoe crab, *Limulus polyphemus* L. *Proc. Nat. Acad. Sci., U.S.*, 71:2558-2562.

Tondo, C.V., J. Bonaventura, C. Bonaventura, M. Brunori, G. Amiconi and E. Antonini. Functional properties of hemoglobin Porto Alegre ($alpha_2^A beta_2^9$ ser->Cys) and the reactivity of its extra cysteinyl residue. *Biochim. Biophys. Acta*, 342:15-20.

1975

Bonaventura, C., J. Bonaventura, G. Amiconi, L. Tentori, M. Brunori and E. Antonini. Hemoglobin Abruzzo (beta 143 (H21) His->Arg): Consequences of altering the 2,3-diphosphoglycerate binding site. *J. Biol. Chem.*, 250:6273-6277.

Bonaventura, J., C. Bonaventura, G. Amiconi, L. Tentori, M. Brunori and E. Antonini. Allosteric interactions in non-alpha chains isolated from normal human hemoglobin, fetal hemoglobin and hemoglobin Abruzzo (beta 143 (H21) His->Arg). *J. Biol. Chem.*, 250:6278-6281.

Bonaventura, J., C. Bonaventura and B. Sullivan. Hemoglobins and hemocyanins: comparative aspects of structure and function. *J. Exp. Zool.*, 194:155-174.

Bonaventura, J., C. Bonaventura, B. Sullivan and G. Godette. Hemoglobin Deer Lodge ($beta_2$ His->Arg): Consequences of altering the 2,3-diphosphoglycerate binding site. *J. Biol. Chem.*, 250:9250-9255.

Chiancone, E., J.E. Nome, S. Forsen, J. Bonaventura, M. Brunori, E. Antonini and J. Wyman. Identification of chloride-binding sites in hemoglobin by nuclear-magnetic-resonance quadrupole-relaxation studies of hemoglobin digests. *Eur. J. Biochem.*, 55:385-390.

Mangum, C., B. Woodin, C. Bonaventura, B. Sullivan and J. Bonaventura. The role of coelomic and vascular hemoglobin in the annelid family Terebellidae. *Comp. Biochem. Physiol.*, 51A:281-294.

Sullivan, B., J. Bonaventura, C. Bonaventura, L. Pennell, J. Elliott, R. Boyum and W. Lambie. The structure and evolution of parvalbumins. I. Amino acid compositional studies of parvalbumins from four perciform species. *J. Mol. Evol.*, 5:103-116.

1976

Bonaventura, C., J. Bonaventura, B. Kitto, M. Brunori and E. Antonini. Functional consequences of ligand-linked dissociation in hemoglobin from the sea cucumber *Molpadia arenicola*. *Biochim. Biophys. Acta*, 428:779-786.

Bonaventura, C., B. Sullivan, J. Bonaventura and M. Brunori. Spot hemoglobin: studies on the Root effect hemoglobin of a marine teleost. *J. Biol. Chem.*, 251:1871-1876.

Bonaventura, C., B. Sullivan, J. Bonaventura and M. Brunori. Hemocyanin of the horseshoe crab, *Limulus polyphemus*: a temperature-jump study of the oxygen kinetics of the isolated components. pp. 265-270. In: *Structure and Function of Hemocyanin*. J.V. Bannister, editor. Springer-Verlag, Berlin.

Bonaventura, J., C. Bonaventura and B. Sullivan. Properties of the oxygen binding domains isolated from subtilisin digests of six molluscan hemocyanins. pp. 206-216. In: *Structure and Function of Hemocyanins*, J.V. Bannister, editor. Springer-Verlag, Berlin.

Bonaventura, J., C. Bonaventura, B. Sullivan, G. Ferruzzi, P.R. McCurdy, J. Fox and W.F. Moo-Penn. Hemoglobin Providence: functional consequences of two alterations of the 2,3-diphosphoglycerate binding site at position beta 82. *J. Biol. Chem.*, 251:7563-7571.

Bossa, F., D. Barra, M. Coletta, F. Martini, A. Liverzani, R. Petruzzelli, J. Bonaventura and M. Brunori. Primary structure of hemoglobins from trout (*Salmo irideus*). Partial determination of amino acid sequence of hemoglobin trout IV. *FEBS Lett.*, 64:76-80.

Chiang, S.C., J. Bonaventura, C. Bonaventura, B. Sullivan, F.K. Swchweighardt and N.C. Li. *Limulus polyphemus* hemocyanin: A nuclear magnetic resonance study of its subunits. pp. 144-149. In: *Structure and Function of Hemocyanin*, J.V. Bannister, editor. Springer-Verlag, Berlin.

Moo-Penn, W., D. Jue, K. Bechtel, M. Johnson, R. Schmidt, P. McCurdy, J. Fox, J. Bonaventura, B. Sullivan and C. Bonaventura. Hemoglobin Providence: a human hemoglobin variant occurring in two forms *in vivo*. *J. Biol. Chem.*, 251:7557-7562.

Schutter, W.G., E.F.J. van Bruggen, J. Bonaventura, C. Bonaventura and B. Sullivan. Structure, dissociation and reassembly of *Limulus polyphemus* hemocyanin. pp. 13-21. In: *Structure and Function of Hemocyanin*, J.V. Bannister, editor. Springer-Verlag, Berlin.

Schwantes, A., M.L. Schwantes, C. Bonaventura, B. Sullivan and J. Bonaventura. Hemoglobins of *Boa constrictor amarali*. *Comp. Biochem. Physiol.*, 54B:447-450.

Sullivan, B., J. Bonaventura, C. Bonaventura and G. Godette. Hemocyanin of the horseshoe crab, *Limulus polyphemus*. Structural differentiation of the isolated components. *J. Biol. Chem.*, 251:7644-7648.

Sullivan, B., J. Bonaventura, C. Bonaventura and P.E. Nute. Structure and function of baboon hemoglobins. pp. 277-288. In: *Wennergren Symposium on Progress in Molecular Anthropology*, M. Goodman and R. Tashina, editors. Plenum Press, New York.

Weber, R., B. Sullivan, J. Bonaventura and C. Bonaventura. The hemoglobin system of the primitive fish *Amia calva*: isolation and functional characterization of the individual hemoglobin components. *Biochim. Biophys. Acta*, 434:18-31.

1977

Amiconi, G., C. Bonaventura, J. Bonaventura and E. Antonini. Functional properties of normal and sickle cell hemoglobin in polyethylene glycol 6000. *Biochim. Biophys. Acta*, 495:279-286.

Bonaventura, C., B. Sullivan, J. Bonaventura and S. Bourne. Anion modulation of the negative Bohr effect of hemoglobin from a primitive amphibian. *Nature*, 265:474-476.

Bonaventura, J., C. Bonaventura and B. Sullivan. Non-heme oxygen transport proteins. pp. 177-220. In: *Oxygen and Physiological Function*, F. Jobsis, editor. Professional Information Library, Dallas.

Brouwer, M., C. Bonaventura and J. Bonaventura. Oxygen binding by *Limulus polyphemus* hemocyanin: allosteric modulation by chloride ions. *Biochemistry*, 16:3897-3902.

Moo-Penn, W.F., K.C. Bechtel, R.M. Schmidt, M.H. Johnson, D.L. Jue, D.E. Schmidt, Jr., W.M. Dunlap, S.J. Opella, J. Bonaventura and C. Bonaventura. Hemoglobin Raleigh (beta I Val->Ac-Ala): Structural and functional characterization. *Biochemistry*, 16:4872-4879.

Terwilliger, R., N. Terwilliger, C. Bonaventura and J. Bonaventura. Oxygen binding domains of *Helisoma trivolvis* hemoglobin. *Biochim. Biophys. Acta*, 494:416-425.

Weber, R.E., C. Mangum, H. Steinman, C. Bonaventura, B. Sullivan and J. Bonaventura. Hemoglobins of two terebellid polychaetes: *Enoplobranchus sanguineus* and *Amphitrite ornata*. *Comp. Biochem. Physiol.*, 56A:179-187.

Weber, R.E., B. Sullivan, J. Bonaventura and C. Bonaventura. The haemoglobin systems of the blood worms, *Glycera dibranchiata* and *G. americana*. Oxygen binding properties of haemolysates and component haemoglobins. *Comp. Biochem. Physiol.*, 58B:183-187.

1978

Bonaventura, C. and J. Bonaventura. Anionic control of hemoglobin function. pp. 647-663. In: *Biochemical and Clinical Aspects of Hemoglobin Abnormalities*, W.S. Caughey, editor. Academic Press, New York.

Bonaventura, C., J. Bonaventura, M. Brunori and M. Wilson. Functional studies on crosslinked bovine cytochrome c oxidase. *FEBS Lett.*, 85:30-34.

Bonaventura, J., C. Bonaventura and G. Lapennas. Hemoglobin engineering: consequences of alterations at functionally sensitive sites particularly susceptible to chemical or enzymatic attack. pp. 109-122. In: *Biochemical and Clinical Aspects of Hemoglobin Abnormalities*, W.S. Caughey, editor. Academic Press, New York.

Brouwer, M., C. Bonaventura and J. Bonaventura. Analysis of the effect of three different allosteric ligands on oxygen binding by hemocyanin of the shrimp *Penaeus setiferus*. *Biochemistry*, 17:2148-2154.

Brouwer, M., M. Ryan, J. Bonaventura and C. Bonaventura. Functional and structural properties of *Murex fulvescens* hemocyanin: isolation of two different subunits required for reassociation of a molluscan hemocyanin. *Biochemistry*, 17:2810-2815.

Bucci, E., A. Salahuddin, J. Bonaventura and C. Bonaventura. Characterization of the ionizable groups interacting with anionic allosteric effectors of human hemoglobin. *J. Biol. Chem.*, 253:821-827.

Sasaki, J., T. Imamura, T. Yanase, D. Atha, A. Riggs, J. Bonaventura and C. Bonaventura. Hemoglobin Hirose, a human hemoglobin variant with the substitution at the $alpha_2beta_2$ interface: subunit dissociation and the equilibria and kinetics of ligand binding. *J. Biol. Chem.*, 253:87-94.

Sekino, T., A. Focesi, C. Bonaventura and J. Bonaventura. Functional properties of *Aplysia brasiliana* myoglobin. *Comp. Biochem. Physiol.*, 61A:223-226.

Sekino, T., A. Focesi, C. Bonaventura and J. Bonaventura. Purification and thermal denaturation of *Aplysia brasiliana* myoglobin. *IRCS Medical Science*, 6:261.

Weber, R.E., J. Bonaventura, B. Sullivan and C. Bonaventura. Oxygen equilibria and ligand-binding kinetics of erythrocruorins from two burrowing polychaetes of different modes of life, *Marphysa sanguinea* and *Diopatra cuprea*. *J. Comp. Physiol.*, 123B:177-184.

1979

Atha, D., A. Riggs, J. Bonaventura and C. Bonaventura. 1979. Hemoglobins of the tadpole of the bullfrog, *Rana catesbeiana*: pH dependence of ligand binding and subunit dissociation equilibria and kinetics. *J. Biol. Chem.*, 254:3393-3400.

Bijlholt, M.M.C., E.F.J. van Bruggen and J. Bonaventura. Dissociation and reassembly of *Limulus polyphemus* hemocyanin. *Eur. J. Biochem.*, 95:399-405.

Bonaventura, J., M. Brunori, M.T. Wilson, J. Martin, R.E. Garlick and J. Davis. Properties of hemocyanins isolated from Amazon River arthropods and molluscs. *Comp. Biochem. Physiol.*, 62A:251-256. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):337-345.

Brunori, M., J. Bonaventura, A. Focesi, M.I. Galdames Portus and M.T. Wilson. Separation and characterization of the hemoglobin components of *Pterygoplichthys pardalis*, the Acaridbodo. *Comp. Biochem. Physiol.*, 62A:173-177. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):231-237.

Farmer, M., R. Weber, J. Bonaventura, R. Best and D. Domining. Functional properties of hemoglobin and whole blood in an aquatic mammal, the Amazonian manatee (*Trichechus inunguis*). *Comp. Biochem. Physiol.*, 62A:231-238. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):311-321).

Focesi, A., M. Brunori, J. Bonaventura, M.T. Wilson and M.I. Galdames Portus. Effect of pH on the kinetics of oxygen and carbon monoxide reactions with hemoglobin from the air-breathing fish, *Loricariichthys*. *Comp. Biochem. Physiol.*, 62A:169-171. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):227-230.

Garlick, R., J. Bonaventura, J. Martin, and D. Powers. Functional studies on the single component hemoglobin from an Amazon knife fish, *Sternopygus marcrurus*. *Comp. Biochem. Physiol.*, 62A:201-205. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):267-273).

Lamy, J., Lamy, J., Weill, J., Bonaventura, J., Bonaventura, C., and Brenowitz, M. Immunological correlates between the multiple subunits of *Limulus polyphemus* and *Tachypleus tridentatus*. *Arch. Biochem. Biophys.*, 196:324-339.

Martin, J., J. Bonaventura, M. Brunori, H. Fyhn, U. Fyhn, R. Garlick, D. Powers and M. Wilson. The isolation and characterization of the hemoglobin components of *Mylossoma* sp., an Amazonian teleost. *Comp. Biochem. Physiol.*, 62A:155-162. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):205-217).

Martin, J., J. Bonaventura, M. Brunori, R. Garlick and D. Powers. The root effect hemoglobin of the Jaraqui, a teleost fish, *Prochilodus* sp. *Comp. Biochem. Physiol.*, 62A:195-200. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):259-266).

Martin, J., J. Bonaventura, H. Fyhn, U. Fyhn, R. Garlick and D. Powers. Structural and functional studies of hemoglobins isolated from Amazon stingrays of the genus *Potamotrygon*. *Comp. Biochem. Physiol.*, 62A:131-138. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):173-184).

Terwilliger, N., R.C. Terwilliger, M. Applestein, C. Bonaventura and J. Bonaventura. Subunit structure and oxygen binding by hemocyanin of the isopod *Ligia exotica*. *Biochemistry*, 18:102.

Wilson, M., J. Bonaventura and M. Brunori. Mitochondrial cytochrome content and cytochrome oxidase activity of some Amazonian fish. *Comp. Biochem. Physiol.*, 62A:245-249. (Also published in Portuguese in *Supl. Acta Amazonica* 8(4):331-336).

1980

Bonaventura, C., and Bonaventura, J. Anionic control of function in vertebrate hemoglobins. *Am. Zool.*, 20:131-138.

Bonaventura, C., and J. Bonaventura. Competition in oxygen-linked anion binding to normal and variant human hemoglobins. In: *Abnormal Human Hemoglobins and Red Cell Enzymes*, T. Huisman, Ed. Marcel Dekker, NY. Hemoglobin: 4(3&4):275-289.

Bonaventura, C., J. Bonaventura and M. Brouwer. Assembly processes in oligomers containing structurally distinct subunits: Respiratory proteins as models. *Biophys. J.*, 32:429.

Bonaventura, J., and Bonaventura, C. Hemocyanins: Relationships in their structure, function and assembly. *Am. Zool.*, 20:7-17.

Bonaventura, J., and Bonaventura, C. Respiratory proteins: Molecular interfaces between the organisms and its environment. In: *Animals and Environmental Fitness*. R. Gilles, Ed. Pergamon Press, pp. 157-170.

Bonaventura, J., and S.C. Wood. Respiratory pigments: Overview. *Am. Zool.*, 20:5-6.

Chiancone, E., Ascoli, F., Brenowitz, M., Bonaventura, C., and Bonaventura, J. *Amphitrite ornata* erythrocruorin I. Structural properties and characterization of subunit interactions. *Biochim. Biophys. Acta*, 623:146-162.

Focesi, A., Jr., S.H. Ogo, C. Bonaventura and J. Bonaventura Kinetics of oxygen and carbon monoxide binding to the hemoglobins of the Water Snakes *Liophis miliaris* and *Helicops modestus*. *Comp. Biochem. Physiol.*, 67B:555-559.

Lamy, J., Lamy, J., Bonaventura, J. and Bonaventura, C. Structure, function and assembly in the hemocyanin system of the Scorpion *Androctonus australis*. *Biochemistry,* 19:3033-3039.

Poyart, C., E. Bursaux, A. Arnone, J. Bonaventura, and C. Bonaventura. Structural and functional studies of hemoglobin Suresnes (alpha 1 ^{1} His->Arg): Consequences of disrupting an oxygen-linked anion-binding site. *J. Biol. Chem.*, 255:9465-9473.

van Bruggen, E.F.J., M.M.C. Bijlholt, W.G. Schutter, T. Wichertjes, J. Bonaventura, C. Bonaventura, J. Lamy, J. Lamy, M. Leclerc, H.J. Schneider, J. Markl and B. Linzen. The role of structurally diverse subunits in the assembly of three cheliceratan hemocyanins. *FEBS Lett.*, 116:207-210.

Wilson, M.T., W. Lalla-Maharajh, V. Darley-Usmar, J. Bonaventura, C. Bonaventura and M. Brunori. Structural and functional properties of cytochrome c oxidases isolated from sharks. *J. Biol. Chem.*, 255:2722-2728.

1981

Bonaventura, C., and Bonaventura, J. The nature of allostery in hemocyanins. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function.* Marcel Dekker pp. 677-691.

Bonaventura, C., J. Bonaventura, K.I. Miller and K.E. Van Holde. Hemocyanin of the chambered nautilus: Structure-function relationships. *Arch. Biochem. Biophys.*, 211:589-598.

Bonaventura, J. and C. Bonaventura. Preparation of high molecular weight invertebrate hemoglobins. In: *Methods in Enzymology*, (Vol. 76), Antonini, E., Rossi-Bernardi, L., and Chiancone, E., Eds., Academic Press, NY. pp. 43-54.

Brenowitz, M., C. Bonaventura, J. Bonaventura and E. Gianazza. Subunit composition of a high molecular weight oligomer: *Limulus polyphemus* hemocyanin. *Arch. Biochem. Biophys.*, 210:748-761.

Brouwer, M., C. Bonaventura and J. Bonaventura. Effect of oxygen and allosteric effectors on structural stability of the oligomeric hemocyanins of the arthropod, *Limulus polyphemus*, and the mollusc, Helix pomatia. *Biochemistry*, 20:1842-1848.

Brouwer, M., Bonaventura, C. and Bonaventura, J. Oxygen-linked changes of the quaternary structure of two structurally different hemocyanins. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function.* Marcel Dekker pp. 761-774.

Brunori, M., Kuiper, H.K., Antonini, E., Bonaventura, C., and Bonaventura, J. Kinetics of oxygen binding by hemocyanins. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function.* Marcel Dekker pp. 693-701.

Chiancone, E., G. Ferruzzi, C. Bonaventura and J. Bonaventura. *Amphitrite ornata* erythrocruorin. II. Molecular controls of function. *Biochim. Biophys. Acta*, 670:84-92.

Lamy, J., Lamy, J., Bonaventura, J., and Bonaventura, C. Functional properties of Androctonus australis hemocyanin: Comparison between native hemocyanin and reconstituted molecules from isolated subunits. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function.* Marcel Dekker pp. 785-796.

Lapennas, G.N., J.M. Colacino and J. Bonaventura. Thin-layer methods for determination of oxygen binding curves of hemoglobin solutions and blood. In: *Methods in Enzymology*, (Vol. 76), Antonini, E., Rossi-Bernard, L., and Chiancone, E., Eds., Academic Press, NY. pp. 449-470.

Markl, J., C. Bonaventura and J. Bonaventura. Hemocyanins in Spiders, XIII: Kinetics of oxygen dissociation from individual subunits of *Eurypelma* and *Cupiennius* hemocyanins. *Hoppe-Seylers Z. Physiol. Chem.*, 362: 429-437.

Mills, B.P., G.E. Nichols, R.E. Frank, J. Bonaventura and R. Roxby. Molecular Mass Measurements of the Alkaline Dissociation Products of *Murex fulvescens* Hemocyanin. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function.* Marcel Dekker pp. 117-124.

Senozan, N., J. Landrum, J. Bonaventura and C. Bonaventura. Hemocyanin of the giant keyhole limpet, *Megathura crenulata*. In: Lamy/Lamy: *Invertebrate Oxygen Binding Proteins: Structure, Active Site and Function*. Marcel Dekker pp. 703-717.

Wood, E.J. and J. Bonaventura. Identification of *Limulus polyphemus* haemocyanin messenger RNA. *Biochem. J.*, 196:653-656.

1982

Bickar, D., J. Bonaventura and C. Bonaventura. Cytochrome c oxidase binding of hydrogen peroxide. *Biochemistry*, 21:2661-2666.

Bonaventura, J., C. Bonaventura and M. Brouwer. Effects of Heavy Metals on the Respiratory Proteins of Marine Organisms in Relation to Environmental Pollution. In: *Structural and Funtional Relationships in Biochemical Systems*, F. Bossa, E. Chiancone, A. Finazzi-Agro and R. Strom, eds. Plenum Univ. Press, pp. 75-83.

Brouwer, M., C. Bonaventura, and J. Bonaventura. Heavy metal ion interactions with *Callinectes sapidus* hemocyanin: Structural and functional changes induced by a variety of heavy metal ions. *Biochemistry*, 21:2529-2538.

Brouwer, M., C. Bonaventura, and J. Bonaventura. Chloride and pH dependence of cooperativeinteractions in *Limulus polyphemus* hemocyanin. In: *Physiology and Biochemistry of Horseshoe Crabs*, J. Bonaventura and C. Bonaventura, Eds. A.R. Liss Publishing Co., NY. pp. 231-256.

Farmer, M., C. Bonaventura and J. Bonaventura. Effects of anions and CO on the dissociation of liganded human hemoglobin and human hemoglobin variants. In: *Hemoglobin and Oxygen Binding*, Chien Ho, Ed. Elsevier. pp. 257-261.

Lamy, J., M. LeClerc, A. Cisse, J. Bonaventura and C. Bonaventura. Preliminary results on possible immunological relatedness between molluscan and arthropodan hemocyanins. In: *Structure and Function of Invertebrate Proteins; Life Chemistry Reports*, E.J. Wood, Ed. Harwood Publishers, pp. 143-146.

Lamy, J., P-V. Sizaret, J. Lamy, R. Feldmann, J. Bonaventura and C. Bonaventura. Preliminary report on the quaternary structure of *Limulus polyphemus* hemocyanin. In: *Structure and Function of Invertebrate Proteins; Life Chemistry Reports*, E.J. Wood, Ed. Harwood Publishers, pp. 47-50.

Schroeder, W.A., J.R. Shelton, J.B. Shelton, G. Apell, L. Evans, J. Bonaventura and R.S. Fang. The partial amino acid sequence of human erythrocyte catalase . *Arch. Biochem. Biophys.*, 214:422-424.

Schroeder, W.A., J.R. Shelton, J.B. Shelton, B. Robberson, G. Apell, R.S. Fang, and J. Bonaventura. The complete amino acid sequence of bovine liver catalase[1,2] and the partial sequence of bovine erythrocyte catalase. *Arch. Biochem. Biophys.*, 214:397-421.

1983

Bonaventura, C. and J. Bonaventura. Respiratory pigments: Structure and function. In: *The Mollusca: Environmental Biochemistry and Physiology*, vol. 2. Wilbur, K., Ed. Academic Press, pp. 1-50.

Bonaventura, J., and C. Bonaventura. Flatfish, fireflies and sharks: Behavior modification induced by natural repellants. In: *Shark Repellants from the Sea: New Perspectives*, Bernard J. Zahuranec, Ed., AAAS Symposium Volume, pp. 115-133.

Brenowtiz, M., C. Bonaventura and J. Bonaventura. Assembly and Calcium-induced cooperativity of *Limulus* IV Hemocyanin: A Model System for Analysis of Structure-Function Relationships in the Absence of Subunit Heterogeneity. *Biochemistry*, 22:4707-4713.

Brenowitz, M., Bonaventura, J., and Bonaventura, C. Chapter 9.8; Haemocyanins. In: *Electrophoresis*, Journal of Chromatography Library, Vol. I8B, Z. Deyl, Ed., Elsevier, pp. 156-160.

Brouwer, M., C. Bonaventura and J. Bonaventura. Metal ion interactions with *Limulus polyphemus* and *Callinectes sapidus* hemocyanin: Stoichiometry and structural and functional consequences of calcium (II), cadmium (II), zInc (II) and mercury (II) binding. *Biochemistry*, 22:4713-4723.

Greenaway, P., J. Bonaventura and H.H. Taylor. Aquatic gas exchange in the freshwater/land crab *Holthuisana transversa*. *J. Exper. Biol.*, 103:225-236.

Greenaway, P., H.H. Taylor and J. Bonaventura. Aerial gas exchange in Australian freshwater/land crabs of the genus *Holthuisana*. *J. Exper. Biol.*, 103:237-251.

Lamy, J., J. Lamy, P.-Y. Sizaret, P. Billiald, P. Jolles, J. Jolles, R.J. Feldmann, J. Bonaventura. The quaternary structure of *Limulus polyphemus* hemocyanin. *Biochemistry*, 22:5573-5583.

Lamy, J., P.-Y. Sizaret, J. Frank, A. Verschoor, R. Feldmann, and J. Bonaventura. Architecture of *Limulus polyphemus* hemocyanin. *Biochemistry*, 21:6825-6833.

Primor, N., C. Bonaventura and J. Bonaventura. Effects of Moses Sole secretion and its active factor, pardaxin, on elasmobranchs. In: *Shark Repellants from the Sea: New Perspectives*, Bernard J. Zahuranec, Ed. AAAS Symposium Volume, pp. 151-156.

1984

Bickar, D., C. Bonaventura and J. Bonaventura. Carbon monoxide-driven reduction of heme proteins. *J. Biol. Chem.*, 259:10777-10783.

Bickar, D., J. Bonaventura, C. Bonaventura, H. Auer and M. Wilson. The paradoxical effects of methyl mercury on the kinetics of cytochrome c oxidase. *Biochemistry*, 23:680-684.

Bonaventura, C., J. Bonaventura, I.R. Hooper and T. Marshall. Underwater life support based on immobilized oxygen carriers. *Appl. Biochem. Biotech.*, 9:65-80.

Brenowitz, M., C. Bonaventura, and J. Bonaventura. Comparison of the physical and functional properties of the 48-subunit native molecule and the 24-, and 12-subunit dissociation intermediates of *Limulus polyphemus* hemocyanin. *Biochemistry*, 23:879-888.

Brenowtiz, M., C. Bonaventura, and J. Bonaventura. Self-association and oxygen-binding characteristics of the isolated subunits of *Limulus polyphemus* hemocyanin. *Arch. Biochem. Biophys.*, 230:238-249.

Johnson, B.A., C. Bonaventura and J. Bonaventura. Allosteric modulation of *Callinectes sapidus* hemocyanin by binding of L-lactate. *Biochemistry*, 23:872-878.

Shih, T., R.T. Jones, J. Bonaventura, C. Bonaventura and R.G. Schneider. Involvement of His HC3(146)beta in hemoglobin's alkaline Bohr effect: Studies of native and N-ethylmaleimide-treated HbA and Hb Cowtown (beta I46 His->Leu). *J. Biol. Chem.*, 259:967-974.

1985

Bickar, D., A. Lehninger, M. Brunori, J. Bonaventura, and C. Bonaventura. Functional equivalence of monomeric (shark) and dimeric (bovine) cytochrome c oxidase. *J. Inorg. Biochem.*, 23:365-372.

Bonaventura, J., and C. Bonaventura. Physiological adaptations and subunit diversity in hemocyanins. In: *Respiratory Pigments in Animals; Relation Structure-Function*, J. Lamy, J.P. Truchot, R. Gilles, Eds. Springer-Verlag, pp. 21-34.

Brunori, M., D. Bickar, J. Bonaventura, and C. Bonaventura. Kinetics of reduction of cytochrome c oxidase by dithionite and the effect of hydrogen peroxide. *J. Biol. Chem.*, 260:7165-7167.

Kempter, B., J. Markl, M. Brenowitz, C. Bonaventura, and J. Bonaventura. Immunological correspondence between arthropod hemocyanin subunits: II. Xiphosuran (*Limulus*) and Spider (*Eurypelma, Cupiennius*) hemocyanin #I#. *Biol. Chem. Hoppe-Seyler*, 366:77-86.

Sugihara, J., T. Imamura, S. Nagafuchi, J. Bonaventura, C. Bonaventura and R. Cashon. Hemoglobin Rahere, a human hemoglobin variant with amino acid substitution at the 2,3-diphosphogly-cerate binding site. Functional consequences of the alteration and effects of bezafibrate on the oxygen bindings. *J. Clin. Invest.*, 76:1169-1173.

Terwilliger, R., N. Terwilliger, C. Bonaventura, J. Bonaventura, and E. Shabtach. Structural and functional properties of hemoglobin from the vestimentiferan pogonophora, *Lamellibrachia. Biochem. Biophys. Acta*, 829:27-33.

1986

Alayash, A.I., A. Dafallah, H. Al-Husayni, A.K. Al-Ali, A. Al-Quorain, A.H.S. Omer, M.T. Wilson, J. Bonaventura, and R. Cashon. Glycosylated hemoglobin levels in a benign form of sickle cell anemia in Saudi Arabia. *Acta Haematologica*, 75:160-164.

Brenowitz, M., K. Munger, C. Bonaventura, and J. Bonaventura. Immunochemical relationships and subunit composition of selected molluscan hemocyanins. In: *Invertebrate Oxygen Carriers*, Linzen, B., Ed. Springer-Verlag, pp. 241-244.

Brouwer, M., D.W. Engel, C. Bonaventura and J. Bonaventura. Toxic trace metals and trace metal binding proteins in marine organisms: An overview. In: *Biology of Benthic Marine Organisms*, Thompson, M.F., R. Sarojini, and R. Nagabhushanam, Eds. Oxford & IBH Publishing Co., New Delhi, pp. 97-109.

Brunori, M., B. Giardina, J. Bonaventura, and C. Bonaventura. Proteine respiratorie di organismimarini e possibili applicazioni biotecnologiche. In: *Proceedings of Marine Biotechnology Symposium*, Italy, pp. 3-11.

Cannon, G.W., M. Tsuchiya, D. Rittschof, and J. Bonaventura. Magnesium dependence of endotoxin-induced degranulation of *Limulus* amebocytes. *Biol. Bull.*, 171:330-337.

Cashon, R., C. Bonaventura, J. Bonaventura, and A. Focesi. The nicotinamide adenine dinucleotides as allosteric effectors of human hemoglobin. *J. Biol. Chem.*, 261:12700-12705.

Fushitani, K., J. Bonaventura, and C. Bonaventura. Isolation of polypeptide chains with heme from the extracellular hemoglobin of *Amphitrite ornata* (Polychaeta, Annelida). *Comp. Biochem. Physiol.*, 84B:137-141.

Rittschof, D., and J. Bonaventura. Macromolecular cues in marine systems. *J. Chem. Ecol.*, 12(5):1013-1023.

Topham, R.W., S. Tesh, C. Bonaventura, and J. Bonaventura. Active-site heterogeneity as revealed by peroxide and mercury: Interactions with purified subunits of Limulus hemocyanin. In: *Invertebrate Oxygen Carriers*, Linzen, B., Ed. Springer-Verlag, pp. 407-416.

1987

Crumbliss, A.L., J.M. Garrison, C.R. Bock, A. Schaaf, C.J. Bonaventura, and J. Bonaventura. Synthesis and characterization of iron (III) chelating analogues of siderophores on organic solid supports. *Inorg. Chim. Acta*, 133:281-287.

Johnson, B.A., J. Bonaventura, and C. Bonaventura. Determination of L-lactate binding stoichiometry and differences in allosteric interactions of structurally distinct homohexamers from *Panulirus interruptus* hemocyanin. *Biochim. Biophys. Acta*, 916:376-380.

Ogo, S.H., A. Focesi, Jr., R. Cashon, C. Bonaventura, and J. Bonaventura. Fluorescence studies on the binding of reduced nicotinamide adenine dinucleotide phosphate to human hemoglobin A and its variant Hemoglobin Providence. *Brazilian J. Med. Biol. Res.*, 20:755-758.

Vinson, C.R., and J. Bonaventura. Structure and oxygen equilibrium of the three coelomic cell hemoglobins of the Echiuran worm *Thalassema mellita* (Conn). *Comp. Biochem. Physiol.*, 87B:361-366.

1988

Alayash, A.I., A.M. El-Hassan, R. Omer, and J. Bonaventura. Glycosylated haemoglobin: An Indicator of Long-Term Blood Glucose in Domestic Sheep and Goats. *Comp. Biochem. Physiol.*, 90A(2):229-231.

Johnson, B.A., C. Bonaventura, and J. Bonaventura. Allostery in *Callinectes sapidus* Hemocyanin: Cooperative Oxygen Binding and Interactions with L-Lactate, Calcium and Protons. *Biochemistry*, 27:1995-2001.

Topham, R., B. Cooper, S. Tesh, G. Godette, C. Bonaventura, and J. Bonaventura. Isolation, purification, and characterization of an iron-binding protein from the horseshoe crab (*Limulus polyphemus*). *Biochem. J.*, 252:151-157.

Topham, R.W., S. Tesh, C. Bonaventura, and J. Bonaventura. Active-Site Heterogeneity in *Limulus* Hemocyanin as Revealed by Reaction with Peroxides. *Arch. Biochem. Biophys.*, 261:299-311.

1989

Alayash, A.I., A. Al-Quorain, and J. Bonaventura. A benign Sickle Cell Disease in a Saudi subject with B^0-Thalassemia and glucose-6-phosphate dehydrogenase deficiency. *Hum. Hered.*, 39:118-120.

Alayash, A.I. and J. Bonaventura. The biochemical basis of sickle cell anemia in Saudi Arabia: A model case of genetic and molecular disorders. *Biochem. Educ.*, 17(1):2-5.

Alayash, A.I., R. Cashon and J. Bonaventura. Modification of the intraerythrocytic hemoglobin S affinity for oxygen by a zinc chelator. *Ann. N.Y. Acad. Sci.*, 565:360-362.

Alayash, A.I., R.E. Cashon, J. Bonaventura, A. Al-Quorain and A.H.S. Omer. Whole blood oxygen equilibrium curves in clinically mild sickle cell anemia patients. *Med. Sci. Res.*, 17:299.

Ogo, S., A. Focesi, Jr., R. Cashon, J. Bonaventura and C. Bonaventura. Interactions of nicotinamide adenine dinucleotides with varied states and forms of hemoglobin. *J. Biol. Chem.*, 264:11302-11306.

1990

Brouwer, M., R. Cashon, and J. Bonaventura. Functional properties of hemoglobin immobilized in coacervates prepared from gelatin A and polyanionic carbohydrates. *Biotech. Bioeng.*, 35:831-836.

Sugita, H., S. Tesh, C. Bonaventura, and J. Bonaventura. Active-Site heterogeneity in Asian horseshoe crab hemocyanins. In: *Proceedings: Invertebrate Dioxygen Carriers*, Leuven University Press, pp. 441-445.

1991

Alayash, A.I., G. Godette, J. Bonaventura, and J. Arnold. Glycosylated hemoglobins in sharks: Relevance to long-term glucose levels and erythrocyte survival. *Comp. Biochem. Physiol.*, 99C:247-249.

Boffi, A., C. Bonaventura, J. Bonaventura, R. Cashon and E. Chiancone. Oxidized dimeric *Scapharca inaequivalvis*. CO-driven perturbation of the redox equilibrium. *J. Biol. Chem.*, 266:17898-17903.

Bonaventura, C., R. Cashon, J. Bonaventura, D. Shih, and M. Perutz. Involvement of the distal histidine in the low affinity exhibited by Hb Chico (Lys E10(66)ß_Thr) and its isolated ß chains. *J. Biol. Chem.*, 266:23033-23040.

Bonaventura, J. In Memorium: An Homage to Robert C. Terwilliger. In: *Proceedings of the Whistler Symposium*, Springer Verlag, NY. pp. xv-xviii.

Burchard, R.P., D. Rittschof, and J. Bonaventura. Adhesion and motility of gliding bacteria on substrata of different surface free energies. *Appl. Environ. Microbiol.*, 56(8):2529-2534.

Rittschof, D., A.R. Schmidt, I.R. Hooper, D.J. Gerhart, D. Gunster, and J. Bonaventura. Molecular mediation of settlement by selected invertebrate larvae. In: *Bioactive Compounds from Marine Organisms*, M.F. Thompson, R. Sarojini, and R. Nagabushanam, Eds. Oxford and IBH Publ. Co., New Delhi, pp. 317-330.

1992

Alayash, A.I., B.A. Brockner Ryan, J.C. Fratantoni, J. Bonaventura, and C. Bonaventura. Hemoglobin-based oxygen carriers (HBOCs): Structural alterations that affect free radical generation. *Biomater. Artif. Cells Immobization Biotechnol.*, 20:277-281.

Alayash, A.I., J.C. Fratantoni, C. Bonaventura, J. Bonaventura, and E. Bucci. Consequences of chemical modifications on the free radical reactions of human hemoglobin. *Arch. Biochem. Biophys.*, 298:114-120.

Brouwer, M., R. Cashon, and J. Bonaventura. Carbodiimide-mediated coupling of benzenepentacarboxylate to human hemoglobin: Structural and functional consequences. *Biomater. Artif. Cells Immobilization Biotechnol.*, 20:323-326.

Brouwer, M., D.W. Engel, J. Bonaventura, and G. Allan Johnson. In vivo magnetic resonance imaging of the blue crab, *Callinectes sapidus*: Effect of cadmium accumulation in tissues on proton relaxation properties. *J. Exp. Zool.*, 263:32-40.

Dewhirst, M.W., E.T. Ong., D. Madwed, B. Klitzman, T. Secomb, D. Brizel, J. Bonaventura, G. Rosner, B. Kavanaugh, J. Edwards and J. Gross. Effects of the calcium channel blocker, flunarizine on tumor microvascular hemodynamics and oxygenation. *Radiat. Res.*, 132:61-68.

Price, R.R., M. Patchan, D. Rittschof, A.S. Clare, and J. Bonaventura. Performance enhancement of natural antifouling compounds and their analogs through microencapsulation and controlled release. *Biofouling,* 6:207-216.

Rittschof, D., A.S. Clare, D.J. Gerhart, Sister Avelin Mary, and J. Bonaventura. Barnacle *in vitro* assays for biologically active substances: Toxicity and settlement inhibition assays using mass cultured *Balanus amphitrite amphitrite* Darwin. *Biofouling*, 6:115-122.

Rittschof, D., A.S. Clare, D.J. Gerhart, J. Bonaventura, C. Smith and M. Hatfield. Rapid field assessment of antifouling and foul-release coatings. *Biofouling*, 6:181-192.

1993

Alayash, A.I., J.C. Fratantoni, C. Bonaventura, J. Bonaventura, and R.E. Cashon. Nitric oxide binding to human ferrihemoglobins cross-linked between either alpha or beta subunits. *Arch. Biochem. Biophys.*, 303:332-338.

Hazes, B., K.A. Magnus, C. Bonaventura, J. Bonaventura, Z. Dauter, K.H. Kalk, and W.G.J. Hol. Crystal structure of deoxygenated *Limlulus polyphemus* subunit II hemocyanin at 2.18 A^{O} resolution: clues for a mechanism for allosteric regulation. *Protein Sci.*, 2:597-619.

1994

Bonaventura, C., M. Arumugam, R. Cashon, J. Bonaventura, and W.F. Moo-Penn. Chloride masks effects of opposing positive charges in Hb A and Hb Hinsdale (ß139 Asn_Lys) that can modulate cooperativity as well as oxygen affinity. *J. Mol. Biol.*, 239:561-568.

Bonaventura, C., J. Bonaventura, R. Stevens, and D. Millington. Acrylamide in polyacrylamide gels can modify proteins during electorphoresis. *Anal. Biochem.*, 222(1):44-48.

Bonilla, G.O., A. Focesi, Jr., C. Bonaventura, J. Bonaventura, and R. Cashon. Functional properties of the hemoglobin from the South American snake *Mastigodryas bifossatus. Comp. Biochem. Physiol., Physiol. A,* 109(4):1085-1095.

Kilbourn, R.G., G. Joly, B. Cashon, J. DeAngelo, and J. Bonaventura. Cell-free hemoglobin reverses the endotoxin-mediated hyporesponsibity of rat aorticrings to α-adrenergic agents. *Biochem. Biophys. Res. Comm.*, 199:155-162.

Magnus, K.A., B. Hazes, H. Ton-That, C. Bonaventura, J. Bonaventura, and W.G.J. Hol. Crystallographic analysis of oxygenated and deoxygenated states of arthropod hemocyanin. *Proteins,* 19:302-309.

Price, R., M. Patchan, D. Rittschof, A. Clare, and J. Bonaventura. Elimination of rosin from vinyl antifouling coatings by the use of novel cylindrical channel forming microtubules. *Trans. Inst. Mar. Engn.*

Rittschof, D., N. Sasikumar, D. Murliess, A.S. Clare, D. Gerhart, and J. Bonaventura. Mixture interactions of lactones, furans and a commercial biocide: toxicity and antibarnacle settlement activity. In: *Recent Developments in Biofouling Control*, M.-F. Thompson, R. Nagabhushanam, R. Sarojini, and M. Fingerman (eds.), pp. 321-334. Oxford and IBH Publ. Co., New Delhi.

Robertson, J.D., J. Bonaventura, and A.P. Kohm. Nitric oxide is required for tactile learning in *Octopus vulgaris. Proc. R. Soc. B Biol. (London)*, 256(1347):269-273..

Stevens, R.D., J. Bonaventura, C. Bonventura, T. R. Fennell, and D.S. Millington. Application of electrospray ionization mass spectrometry for analysis of haemoglobin adducts with acrylonitrile. *Biochem. Soc. Trans., U.K.*, 22:543-547.

1995

Bonaventura, C., J. Bonaventura, and D.F. Bodishbaugh. Environmental bioremediation: Applications and new horizons. In: *Ecotoxicity and Human Health: A Biological Approach to Environmental Remediation*, Bloom, A.D. and F.J. de Serres, Eds. Lewis Publishers. pp. 211-242.

Bonaventura, C., J. Bonaventura, D.F. Bodishbaugh. Environmental bioremediation: Approaches and processes. In: *Ecotoxicity and Human Health: A Biological Approach to Environmental Remediation*, Bloom, A.D. and F.J. de Serres, Eds. Lewis Publishers. pp. 183-210.

Holm, D.E., G. Godette, J. Bonaventura, C. Bonaventura, and J. Peterson. The site ofthe redox-linked proton pump in eukaryotic cytochrome c oxidases. *FEBS Lett.*, 370:53-58.

Meyer, R.E., S. Shan, J. DeAngelo, R.K. Dodge, J. Bonaventura, E.T. Ong, and M.W. Dewhirst. Nitric oxide synthase inhibition irreversilby decreases perfusion in the R3230Ac rat mammary adenocarcinoma. *Br. J. Cancer*, 71(6):1169-1174.

Vomela, J., V. Valek, J. Bonaventura, and R. Kostrica. Esophageal perforation at the thoracoabdominal juncture. [Czech.] *Bratisl. Lek. Listy*, 96(5):274-279.

Weinberg, J.B., E. Doty, J. Bonaventura, and A.F. Haney. Nitric oxide inhibition of human sperm motility. *Fertil. Steril.*, 64(2):408-413.

1996

Jia, L., C. Bonaventura, J. Bonaventura, and J.S. Stamler. S-Nitrosohemoglobin: a dynamic activity and transport function of blood. *Nature* 380:221-226.

[Note also: Perutz, M. 1996. Taking the pressure off. *Nature (News and Views)* 380:205-206.**]**

Mylvaganam, S.E., C. Bonaventura, J. Bonaventura, and E.D. Getzoff. Structural basis for the Root effect in haemoglobin. *Nature Structural Biology* 3:275-283.

[Note also: (1.) Perutz, M. 1996. Cause of the root effect in fish haemoglobins. *Nature Structural Biology (News and Views)* 3:211-212. **(2.)** Howlett, R. (Assoc. Ed., *Nature*). 1996. Root cause of fish buoyancy. *Nature (News and Views).* **]**

1997

Hahn, J.S., R.D. Braun, M.W. Dewhirst, S. Shan, S.A. Snyder, J.M. Taube, E.T. Ong, G.L. Rosner, R.K. Dodge, J. Bonaventura, C. Bonaventura, J. DeAngelo and R.E. Meyer. Stroma-free human hemoglobin A selectively decreases R3230Ac rat mammary adenocarcinoma blood flow and oxygen partial pressure. *Rad. Res.* 147(2):185-194

1998

Bickar, D., C. Bonaventura, and J. Bonaventura. The kinetics of peroxide binding by cytochrome c oxidase. *Biochemistry* (Submitted).

Bonaventura, C., J. Bonaventura, R. Stevens, and D. Millington. Polyacrylamide gel electrophoresis can produce acrylamide adducts. *Protein Sci.* (Submitted).

Bonaventura, C., G. Godette, S. Tesh, D.E. Holm, J. Bonaventura, A.L. Crumbliss, L.L. Pearce and J. Peterson. Internal electron transfer reactions in the carbon monoxide-driven reduction of methemoglobin-copper complexes and cytochrome *c* oxidase. *Biochemistry* (Submitted).

Holm, D.E., G. Godette, C. Bonaventura, J. Bonaventura, M.D. Boatright, L.L. Pearce, and J. Peterson. A carbon monoxide irreducible form of cytochrome c oxidase and other unusual properties of the "monomeric" shark enzyme. *Comp. Biochem. Physiol.* (Submitted).

Holm, D.E., G. Godette, J. Bonaventura, C. Bonaventura, and J. Peterson. Functional and spectral properties of "monomeric" (shark) cytochrome *c* oxidases: evidence for two distinct "resting" forms of the enzyme. *Biochemistry* (Submitted).

Stamler, J.S., L. Jia, J. Eu, J. McMahon, I.T. Demchenko, J. Bonaventura, K. Gernert and C.A. Piantadosi. 1997. Blood flow regulation by S-nitroso hemoglobin in the physiological oxygen gradient. *Science* 276:2034-2037.

2. **Books**:

Physiology and Biochemistry of Horseshoe Crabs, J. Bonaventura and C. Bonaventura, Eds. A.R. Liss Publishing Co., NY. pp. 231-256. (1982).

3.a. **Published Reviews**:

Bonaventura, J. (1978). Review of *Animal Biochromes and Structural Colours*, by D.L. Fox. Physiol. Zool., 5I:IO4.

Bonaventura, J. (1981). Review of *The Chromatography of Hemoglobin (Clinical and Biochemical Analysis Series).* Volume 9. By Walter A. Schroeder and Titus H.J. Huisman. Marcel Dekker, Inc., NY (1980). J. Am. Chem. Soc. 103:7040.

Bonaventura, J. (1989) Review of *Biomedical Importance of Marine Organisms*, Daphne G. Fautin, ed., California Academy of Sciences, San Francisco (1988). *BioScience,* 39(5):332-334.

3.b. **Selected Abstracts**:

1968

Bonaventura, J. and A. Riggs. Isolation of cysteinyl peptides from polymeric hemoglobins of the wild house mouse *Mus musculus* diffuse single alleles. (Abstract). *Genetics,* 60(1 part 2), p. 163.

1969

Kitto, G.B., and J. Bonaventura. Deoxygenation induced aggregation of some invertebrate hemoglobins. (Abstract). *Fed. Proc.*, 28(2):867.

1972

Bonaventura, J., C. Bonaventura, M. Brunori, G. Amiconi, E. Chiancone and E. Antonini. Functional properties of hemoglobin Abruzzo. (Abstract). XIV International Congress of Hematology. Sao Paulo.

4. a. Pat nts

U.S. Patent No. 4,340,587: (Issued 7/20/82)	Repellant Composition and Method of Use.
U.S. Patent No. 4,343,715: (Issued 8/10/82)	Immobilized Hemoglobin, and Processes for Extracting Oxygen from Fluids Using the Same.
U.S. Patent No. 4,427,416: (Issued 1/24/84)	Processes for Extracting Oxygen from Fluids Using Immobilized Hemoglobin.
U.S. Patent No. 4,490,360: (Issued 12/25/84)	Firefly Derived Repellant Compositions and Methods of Use.
U.S. Patent No. 4,602,987: (Issued 7/29/86)	System for the Extraction and Utilization of Oxygen From Fluids.
U.S. Patent No. 4,609,383: (Issued 9/2/86); ***and*** European Patent No. 92202007.8 (Issued 8/25/92)	Apparatus and Method for Extracting Oxygen from Fluids.
U.S. Patent No. 4,629,544 (Issued 12-16-86)	Apparatus and Method for Reversibly Removing Ligands from Carriers.
U.S. Patent No. 4,704,286: (Issued 11/3/87)	Gustatory Additive for Fishing Lures.
U.S. Patent No. 4,751,068: (Issued 6/14/88)	A Method for Catalyzing Oxidation/ Reduction Reactions of Simple Molecules.
U.S. Patent No. 4,761,209 (Issued 08/02/88)	System for the Extraction and Utilization of Oxygen from Fluids
U.S. Patent No. 5,082,642 (Issued 01/21/92)	Method for Catalyzing Oxidation/Reduction Reactions of Simple Molecules.
U.S. Patent No. 5,252,630: (Issued 10/12/93)	Antifouling Coating and Method for Using Same.
U.S. Patent No. 5,296,466 (Issued 03/22/94)	Inhibition of Nitric Oxide-mediated Hypotension and Septic Shock with Iron-containing Hemoprotein.
U.S. Patent No. 5,314,932 (Issued 05/24/94)	Antifouling Coating and Method for Using Same.
U.S. Patent No. 5,334,705 (Issued 08/02/94)	Benzenetricarboxylate Derivative-crosslinked Low Oxygen Affinity Hemoglobin.
U.S. Patent No. 5,349,054 (Issued 09/02/94)	Activated Benzenepentacarboxylate-crosslinked Low Oxygen Affinity Hemoglobin.
U.S. Patent No. 5,480,866 (Issued 02/02/96)	Hemoproteins for Inhibition of Nitric Oxide-mediated Hypotension and Septic Shock.

G:\CLIENTFILD\DUK\96-03PA\13
CAE34/WP5.1
DEB/CAE/ach
February 17, 1998

PATENT APPLICATION
DOCKET NO.: DUK96-03pA

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Jonathan S. Stamler
Serial No.: 08/616,371 Group Art Unit: 1811
Filed: March 15, 1996 Examiner: B. Celsa
For: METHODS FOR PRODUCING AND USING S-NITROSOHEMOGLOBINS

CERTIFICATE OF MAILING

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as First Class Mail in an envelope addressed to: Assistant Commissioner for Patents, Washington, D.C. 20231

on 3/12/98 Anita Heckman
Date Signature

Anita Heckman
Typed or printed name of person signing certificate

DECLARATION OF JOSEPH BONAVENTURA, Ph.D., UNDER 37 C.F.R. § 1.132

Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

I, Joseph Bonaventura, Ph.D., of Durham, North Carolina, hereby state that.

1. I am a Professor in the Nicholas School of the Environment and the Department of Cell Biology at Duke University Medical School. I have held academic appointments at Duke University since 1972. A somewhat abbreviated copy of my curriculum vitae is attached to this Declaration as Exhibit A.

I am the author of over ~~eighty~~ 200 refereed journal articles on various aspects of research specifically on hemoglobin proteins.

I have also studied extensively other heme proteins with structures and functions related to those of hemoglobin. My more recent publications include articles describing the synthesis and properties of S-nitrosohemoglobins (SNO hemoglobins).

2. I have studied Example 19 (pages 58-59) and Figures 28-30 of the published PCT application, International Publication Number WO 93/09806. Example 19 is entitled "S Nitrosylation of Hemoglobin Increases Hemoglobin Oxygen Binding."

3. The first paragraph on page 58 is particularly confusing because of a reagent missing from the procedure. According to page 58, line 5 of WO 93/09806, 12.5 μM hemoglobin was reacted, but it is not given in the Example with what reagent hemoglobin is reacted. According to the letter of the method, the hemoglobin just reacted -- maybe with itself? Alternatively, the description might be indicating that the reaction of hemoglobin occurred with the buffer. However, no buffer is given.

If one assumes that the reagent intended to have been inserted on page 58, line 5 is SNOAc (as described below in the Example for a separate set of experiments whose results are indicated as being shown in Figure 29) at the concentrations and conditions given on page 58, lines 4-6, SNO hemoglobin would not be made. Further, the synthesis of SNO-hemoglobin could not be assessed as described. The standard Saville method, as it is described as being used to characterize the results of the "synthesis" process, would only be able to measure concentrations of the reagent SNOAc (S nitroso N acetylcysteine), and not hemoglobin.

Figure 28 is uninterpretable as presented. The abscissa and ordinate are not labeled. It is impossible to tell what was plotted and what the plot is a function of. I can assume that it is SNOAc, because the spectrum is definitely not recognizable as that of hemoglobin or SNO-hemoglobin. If the spectrum were that of SNO-hemoglobin, it would have a very different appearance in

the region of absorbance of light of wavelength 500-600 nm. (Here, I have added the nm units on an assumption based on my inspection of *Figure 29*.)

4. Figure 29 shows a spectral progression of oxy-hemoglobin to met hemoglobin. Thus, it is incorrect to assert, as the text of Example 19 does, that SNOAc does not react in a redox reaction with the metal centers of hemoglobin. Figure 29 shows quite the contrary. Moreover, based on what is given in Example 19, I have no way to assess whether the spectra result from pure hemoglobin alone or some mixture of SNO hemoglobin with something else, or some entirely different alternative. I interpret the spectra as showing that oxy-hemoglobin is being converted to met-hemoglobin.

5. In a further point of confusion, I find the title of Example 19 to be very misleading. It is not explained what is meant by "*increases oxygen binding.*" Is it meant that *more* oxygen molecules bind per hemoglobin molecule? Is it meant that the affinity of hemoglobin for oxygen somehow increases? Nothing is written or shown in Example 19 that is relevant to oxygen binding.

6. Example 19 of PCT application WO 93/09806 describes a putative method for preparing SNO-hemoglobin. At face value the description of the procedure is, at best, difficult to follow. Example 19 does not describe a method that I would be able to follow to successfully synthesize SNO-hemoglobin. In my opinion, no one skilled in the art and science of hemoglobin chemistry would be able to synthesize SNO-hemoglobin based on the description and instructions provided. The points outlined above are hardly the only points that make the description inadequate.

7. I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further, that these

statements are made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Joseph Bonaventura

Joseph Bonaventura, Ph.D.

11 March, 1998

Date